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Different preservatives and dna extraction methods for tissues (Marcia Japonica (clam) tissues in pcr amplification study / Liat Siew Ting.

DIFFERENT PRESERVATIVES AND DNA EXTRACTION METHODS FOR TISSUES OF *MARCIA JAPONICA* (CLAM) IN PCR AMPLIFICATION STUDY

By

Lian Siew Ting

Research Report submitted in partial fulfillment of
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PENGAKUAN DAN PENGESAHAN LAPORAN
PROJEK PENYELIDIKAN I DAN II

Adalah ini diakui dan disahkan bahawa laporan penyelidikan bertajuk: DIFFERENT PRESERVATIVES AND DNA EXTRACTION METHODS FOR TISSUES OF *MARCIA JAPONICA* (CLAM) IN PCR AMPLIFICATION STUDY oleh Lian Siew Ting, no. matrik UK7726 telah diperiksa dan semua pembetulan yang disarankan telah dilakukan. Laporan ini dikemukakan kepada Jabatan Sains Biologi sebagai memenuhi sebahagian daripada keperluan memperolehi Ijazah Sarjana Muda Sains–Sains Biologi, Fakulti Sains dan Teknologi, Kolej Universiti Sains dan Teknologi Malaysia.

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Bp	base pair
C	Cytosine
DNA	Deoxyribonucleic Acid
dNTP	2'- deoxynucleoside-5'- triphosphate (s)
G	Guanine
mM	milimolar
ng	nano gram
OD	Optical Density
PCR	Polymerase Chain Reaction
RAPD	Random Amplified Polymerase DNA
SDS	sodium dodecyl sulfate
TBE	Tris-Borate-EDTA buffer
TNES	Tris-NaCl-EDTA-SDS
VDS	Video Documentation System
w/v	weight/volume
µL	micro liter

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ABSTRACT

Preservation is significant because marine biodiversity is centered in the Indo-Pacific, where immediate DNA analysis is impossible. The genomic DNA that successfully extracted can be used for PCR amplification. The objectives of this study are to measure the DNA purity and quantity from 95% ethanol and TNES-urea buffer and to compare the efficiency of Promega Wizard™ Genomic DNA Purification Kit and phenol-chloroform for DNA extraction . In this experiment, the tissues were preserved in 95% ethanol and TNES-urea buffer. Besides that, the genomic DNA was extracted using the Promega Wizard™ Genomic DNA Purification Kit and Phenol-chloroform. Referring to the results, For the Promega Wizard™ Genomic DNA Purification Kit Extraction, the overall purity of genomic DNA for *Marcia japonica* was in the range of 1.286 (OD_{260}/OD_{280}) and the quantity of genomic DNA was in the range of 1024.58 ng/μl. While for the Phenol-chloroform Extraction, the overall purity of genomic DNA for *M.japonica* was in the range of 1.461 (OD_{260}/OD_{280}) and the quantity of genomic DNA was in the range of 573.33 ng/μL. The genomic DNA of *M. Japonica* have poor purity because the overall purity was in the range of 1.260 to 1.649 (OD_{260}/OD_{280}). The genomic with ratio ranging from 1.8 to 2.0 for PCR requirement in amplification of DNA. Moreover, it was suggested that phenol-chloroform was the best method to extract DNA in this study and TNES-urea buffer was a good preservative to store samples.

PENGAWET DAN KAEDAH PENGEKSTRAKAN DNA YANG BERBEZA UNTUK TISU-TISU *MARCIA JAPONICA* (KEPAH) DALAM KAJIAN AMPLIFIKASI PCR

ABSTRAK

Tujuan kajian ini adalah untuk mengukur ketulenan dan kuantiti DNA daripada tisu yang diekstrak dalam etanol 95% dan TNES-urea buffer serta untuk membandingkan kecekapan antara Promega Wizard™ Genomic DNA Purification Kit dan Fenol-kloroform untuk mengekstrak DNA. Sampel tisu telah diawet dengan menggunakan dua jenis pengawet iaitu etanol 95% dan TNES-urea buffer. Selain itu, genomik DNA telah diekstrak dengan menggunakan kaedah Promega Wizard™ Genomic DNA Purification Kit dan Fenol-kloroform. Mengikut keputusan yang diperolehi, ketulenan genomik DNA bagi *Marcia japonica* yang diekstrak dengan kaedah Promega Wizard™ Genomic DNA Purification Kit adalah 1.286 (OD_{260}/OD_{280}) dan kuantiti bagi genomik DNA adalah 1024.58 ng/µl. Manakala ketulenan bagi tisu yang diekstrak dengan kaedah fenol-kloroform adalah 1.461 dan kuantiti genomik DNA pula adalah 573.33 ng/µl. Genomik DNA bagi *M. japonica* mempunyai ketulenan yang rendah iaitu di antara 1.260 hingga 1.649 (OD_{260}/OD_{280}). Genomik DNA yang mempunyai kadar ketulenan antara 1.8-2.0 adalah sesuai untuk pengekstrakan DNA. Saya mencadangkan fenol-kloroform merupakan kaedah yang paling sesuai untuk pengekstrakan DNA bagi tisu-tisu *M. japonica* di dalam kajian ini.